

AMENDMENTS TO THE CLAIMS

This listing of claims will replace all prior listings of claims in the application.

LISTING OF CLAIMS:

1-15. (Canceled)

16. (Currently Amended) A method for detecting an analyte with an immunoliposome-nucleic acid amplification assay, comprising:
 encapsulating ~~a plurality of~~ 50 to 1,000 identical nucleic acid segments within closed shell liposomal bilayers,
 associating receptors to the extravesicular surface of said liposomal bilayers,
 exposing the receptors to an immobilized target analyte, which binds to the liposomal bilayer associated receptors;
 removing unbound liposomal bilayers;
 lysing the bound liposomal bilayers to release the nucleic acid segments;
 amplifying the nucleic acid segments released from said liposomal bilayers, and
 detecting the amplification products of the nucleic acid segments to quantify the amount of the target analyte.

17. (Previously Presented) The method of claim 16, wherein the target analyte is selected from the group consisting of proteins, nucleic acids, carbohydrates, glycolipids, gangliosides, viruses, bacteria, toxins, chemical warfare agents, explosives, poisons, hormones, cancer-specific soluble biological markers, tumor cell-surface markers, and minor cell components in larger cell populations.

18. (Previously Presented) The method of claim 16, wherein the immunoliposome-nucleic acid amplification assay can be used to spatially localize an analyte within a fresh or fixed tissue section.

19. (Canceled)

20. (Previously Presented) The method of claim 16, wherein the receptors are selected from the group consisting of monoclonal or polyclonal antibodies, antibody Fab' fragments, glycolipids, soluble proteins, dyes, DNA probes, and RNA probes.

21. (Previously Presented) The method of claim 16, comprising anchoring the receptors to the surface of the liposomal bilayers through covalent attachment to a long-chain-length hydrocarbon having 12 to 24 carbons.

22. (Previously Presented) The method of Claim 21, wherein the long-chain-length hydrocarbon comprises carboxylic acids, amines, thiols, alcohols, aldehydes, nitrites, amides, or halides.

23. (Previously Presented) The method of Claim 16, wherein associating receptors to the extravesicular surface of the liposomal bilayers comprises covalently attaching an antibody to glycolipids or phospholipids.

24. (Previously Presented) The method of Claim 16, wherein associating receptors to the extravesicular surface of the liposomal bilayers comprises electrostatically coupling charged receptors to charged lipids in the liposomal bilayers.

25. (Canceled)

26. (Previously Presented) The method of claim 16, comprising anchoring integral membrane protein receptors to the liposomal bilayers by direct incorporation into the liposomal bilayers.

27. (Previously Presented) The method of claim 16, comprising reducing non-specific binding of the liposomal bilayers on an immobilizing substrate by varying the lipid composition of the liposomal bilayers to alter the size of the liposome, the fluidity of the bilayer, or the polarity and charge of the surface of the liposomal bilayer.

28. (Previously Presented) The method of claim 16, further comprising reducing non-specific binding of the liposomal bilayers on an immobilizing substrate by altering the charge density of the surface of the liposomal bilayer.

29. (Previously Presented) The method of claim 16, further comprising reducing non-specific binding of the liposomal bilayers on an immobilizing substrate by attaching polyethylene glycol to the surface of the liposomal bilayer.

30. (Previously Presented) The method of claim 16, further comprising reducing non-specific binding of the liposomal bilayers on an immobilized substrate by varying the length of a spacer arm used to attach the receptors to the liposomal bilayers.

31. (Previously Presented) The method of claim 16, further comprising reducing background DNA or RNA contamination of the assay by adding DNase or RNase to the assay solution, thereby degrading background DNA or RNA.

32. (Currently Amended) The method of claim 16, wherein the liposomal bilayers are lysed using **[[a]]** an alcohol or melittin.

33. (Previously Presented) The method of claim 16, wherein said amplifying comprises polymerase chain reaction, real-time PCR, bDNA or Q-beta replicase methods.

34. (Previously Presented) The method of claim 16, wherein said detecting comprises capillary electrophoresis or spectrophotometric assays using nucleic acid-specific dyes.

35. (Previously Presented) The method of claim 16, wherein said amplifying and detecting are coupled.

36. (Canceled)

37. (Previously Presented) The method of claim 16, wherein the target analyte is detected in subattomolar quantities.

38. (Previously Presented) The method of claim 16, further comprising linking a specific receptor to a liposomal bilayer encapsulating nucleic acid segments having a unique nucleotide length, thereby screening for several target analytes at one time.

39. (Previously Presented) The method of claim 16, comprising detecting toxins in soil, water or air.

40. (Canceled)

41. (Previously Presented) The method of claim 16, comprising detecting target analyte in biological fluids.

42. (Previously Presented) The method of claim 16, wherein the immobilized target analyte is immobilized on magnetic micro-particles or a micro-fabricated device.

43. (Currently Amended) A method for detecting an analyte with an immunoliposome-nucleic acid amplification assay, comprising:

encapsulating a plurality of identical nucleic acid segments within closed shell liposomal bilayers,

incorporating receptors into the outer surface of said liposomal bilayers; and exposing the receptors to a target analyte, causing aggregation of the receptors within the plane of the ~~liposomal~~ liposomal bilayers, wherein the aggregation causes the liposomal bilayers to become unstable leading to spontaneous rupture of the ~~liposomal~~ liposomal bilayers, and release of the nucleic acid segments.

44. (Previously Presented) A method according to Claim 16, wherein encapsulating the plurality of identical nucleic acid segments within closed shell liposomal bilayers comprises mixing phospholipid single-shell vesicles with ethanol and calcium chloride to form phospholipids-nucleic acid segment complexes and dialyzing said complexes.

45. (Previously Presented) A method according to Claim 43, wherein encapsulating the plurality of identical nucleic acid segments within closed shell liposomal bilayers comprises mixing phospholipid single-shell vesicles with ethanol and calcium chloride to form phospholipids-nucleic acid segment complexes and dialyzing said complexes.

U.S. Serial No. 10/759,099

AMENDMENT AFTER FINAL REJECTION UNDER 37 C.F.R. 1.116

Docket No. AFIP 03-16

46. (Previously Presented) The method of claim 16, wherein the receptors are monoclonal or polyclonal antibodies.